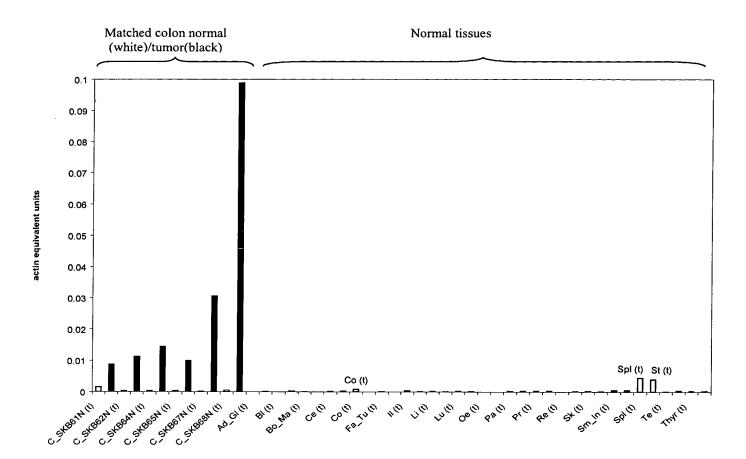
Figure 1: Real-time PCR data using the Taqman probe



THE જ ゃ ✨ ۾ ع 弘太 % જ 00 も **^>** % 0.15 0.05 0.45 0.4 0.35 0.3 0.25 0.2 0.7 actin equivalent units

Figure 2: Real-time PCR expression using Sybr protocol

Figure 3: Coomassie blue stained SDS PAGE of the cell extract from the strain expressing CASB7439

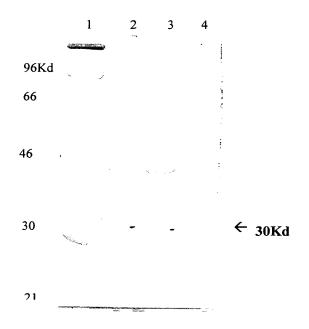


Figure 4: Western blot loaded with cell extract from the strain expressing CASB7439

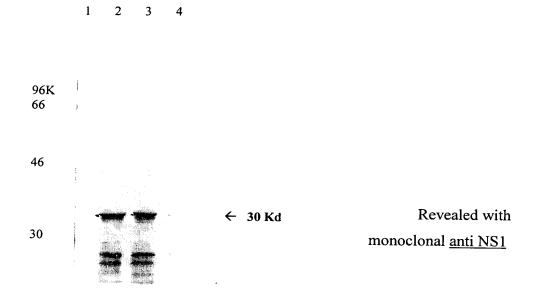


Figure 5: Coomassie-blue stained SDS-PAGE of CASB7439 after purification

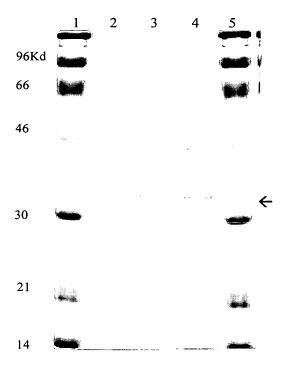
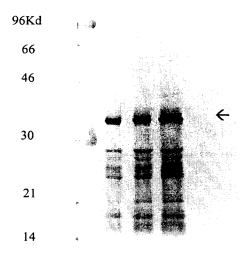


Figure 6: Western blot CASB7439 after purification (monoclonal anti-polyhistidine)

1 2 3 4



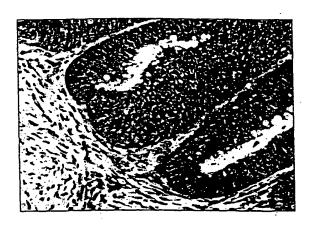


Figure 7
CASB7439 IHC on a Colon Tumour Biopsy

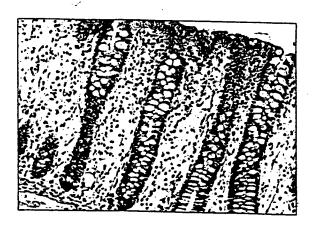


Figure 8
CASB7439 IHC on a Colon Normal Mucosa Biopsy

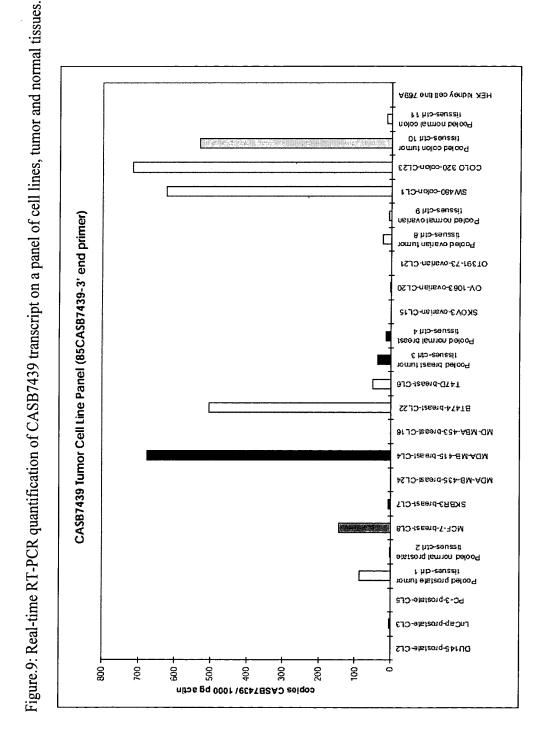


Figure 10: Real-time RT-PCR quantification (3' UTR pimers) of CASB7439 transcript on a panel of tumours grown in SCID mice.

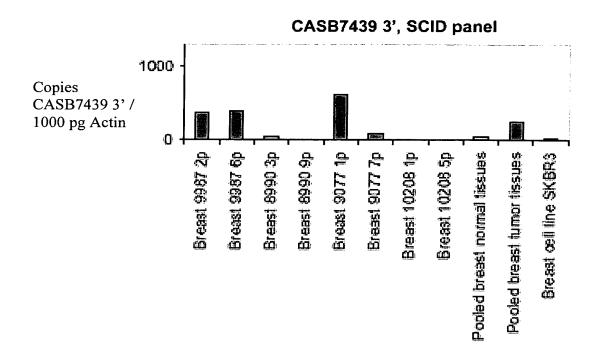


Figure 11: Real-time RT-PCR quantification (ORF primers) of CASB7439 transcript on a panel of tumours grown in SCID mice.

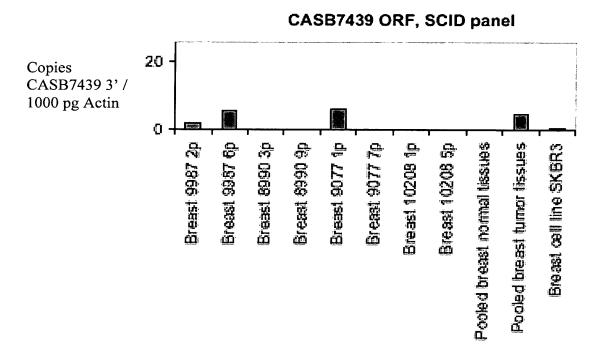


Figure 12: Real-time RT-PCR quantification (3' UTR pimers) of CASB7439 transcript on an extended panel of normal and tumours tissues.



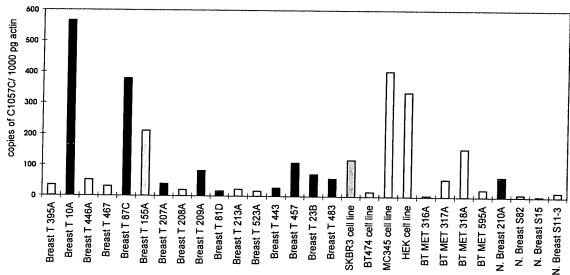


Figure 13: Real-time RT-PCR quantification (3' UTR pimers) of CASB7439 transcript on an extended panel of normal and tumours tissues.

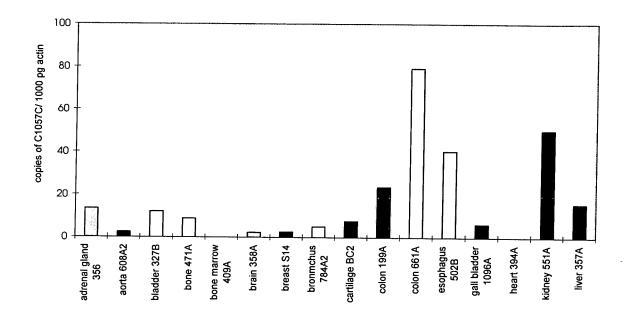


Figure 14: Real-time RT-PCR quantification (3' UTR pimers) of CASB7439 transcript on an extended panel of normal and tumours tissues, continued.

CASB7439 3', Extended Breast Panel

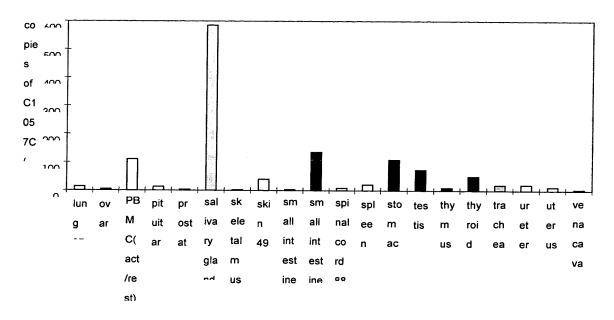


Figure 15: Design of NS1-CASB7439 protein.

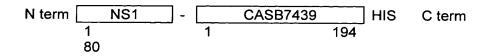


Figure 16: Schematic represention of pMG1 cloning strategy (constructs 1 and 2).

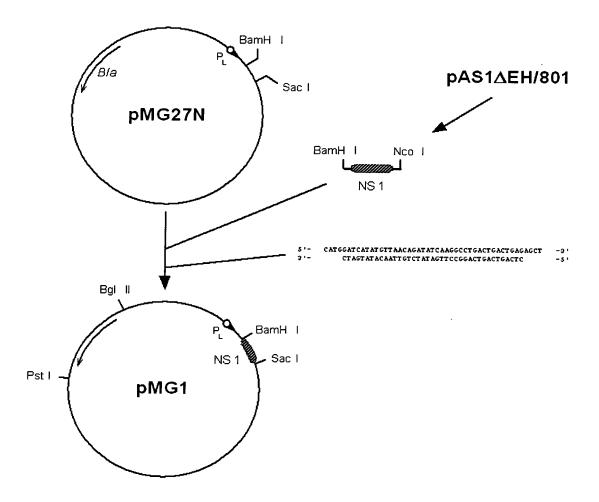
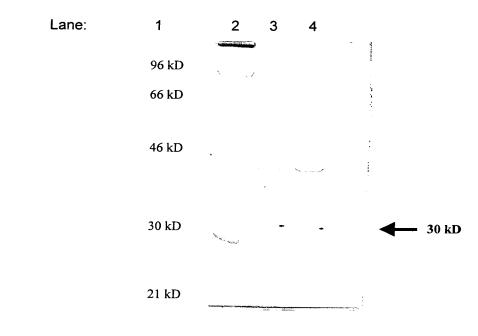


Figure 17: Coomassie blue stained SDS PAGE of the cell extract from the strain expressing CASB7439 construct 1.



5

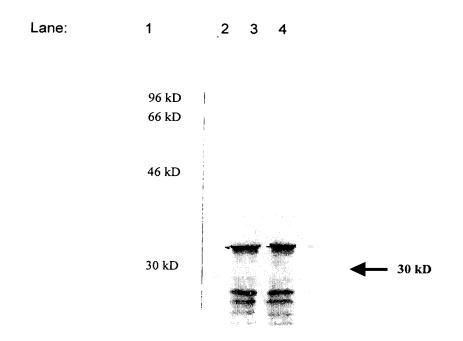
Lane 1: Molecular weights

Lane 2: Whole cellular extract

10 Lane 3: Supernatant of cellular extract

Lane 4: Pellet of cellular extract

Figure 18: Western blot loaded with cell extract from the strain expressing CASB7439 construct 1.



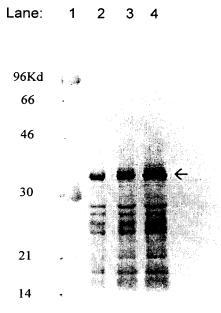
Lane 1: Molecular weights

Lane 2: Whole cellular extract

Lane 3: Supernatant of cellular extract

Lane 4: Pellet of cellular extract

Figure 19: Western blot CASB7439 construct 1 after purification (monoclonal anti-NS1 N terminal fragment).



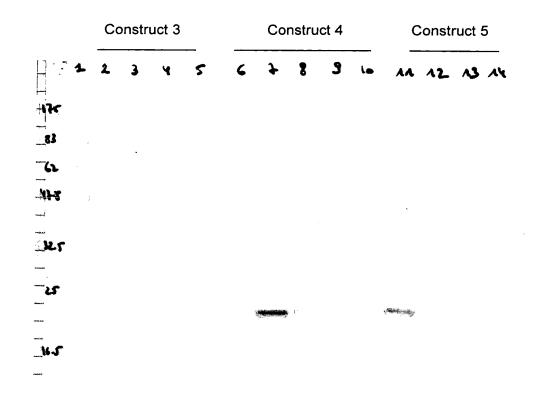
Lane 1: Molecular weights

Lane 2: 2 µl of purified protein

Lane 3: 4 µl of purified protein

Lane 4: 6 µl of purified protein

Figure 20: Comparison of Constructs 3 to 5 expression yields. Western blot on CASB7439 constructs 3 to 5 expressed in *E. coli*, revealed with antipeptide SB600 antibody.



Lane 1: MW; Kd

Lane 2: Construct 2, undiluted.

Lane 3: Construct 2, 1/10 dilution.

Lane 4: Construct 2, 1/50 dilution.

Lane 5: Construct 2, 1/100 dilution.

Lane 6: Construct 3, undiluted.

Lane 7: Construct 3, undiluted.

Lane 8: Construct 3, 1/10 dilution.

Lane 9: Construct 3, 1/50 dilution.

Lane 10: Construct 3, 1/100 dilution.

Lane 11: Construct 4, undiluted.

Lane 12: Construct 4, 1/10 dilution.

Lane 13: Construct 4, 1/50 dilution.

Lane 14: Construct 4, 1/100 dilution.

Figure 21: Expression of C7439 in HEK 293T Cells by DNA Vaccine Vector pJB76 (pJB16/C7439)

Expression of C7439 in HEK 293T Cells by DNA Vaccine Vector pJB76 (pJB16/C7439)

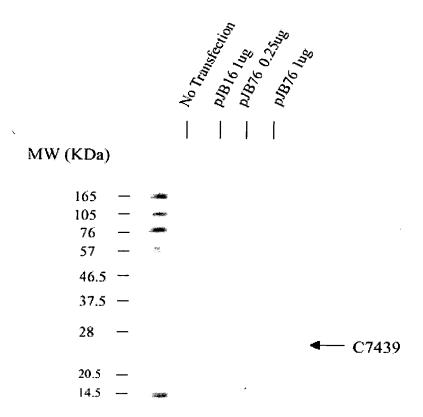
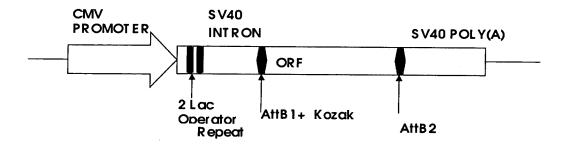


Figure 22: Schematic representation of CASB7439 expressed in a adenoviral live vector

Adenovirus Lac/GW-1 Expression Cassette



AttB1 ACAAGTTT atacaaaAAAGC AttB2 ACCCAGCTTT cttatacAAA

Figure 23: Expression of C7439 in Human Fibroblasts by Recombinant Adenovirus AdC7439

Expression of C7439 in Human Fibroblasts by the Recombinant Adenovirus AdC7439

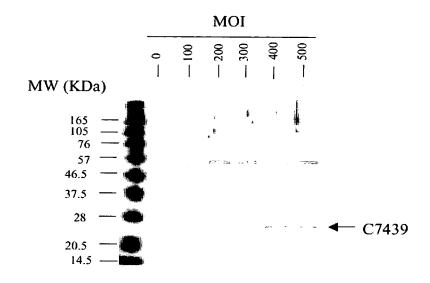


Figure 24: Real-time RT-PCR quantification of CASB7439 transcript on a population of preneoplasic lesions of lung (PLL) patients.

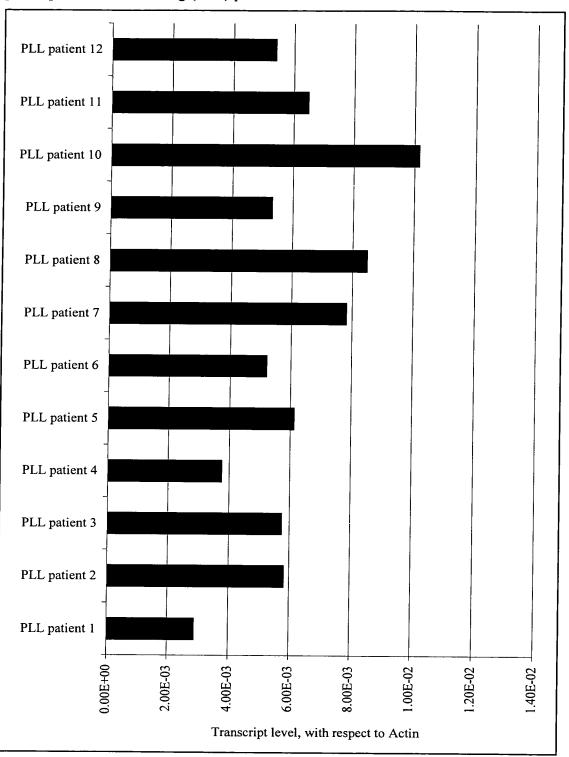


Figure 25: Real-time RT-PCR quantification of CASB7439 transcript on a population oc early stage lung cancer (ESLC) patients.

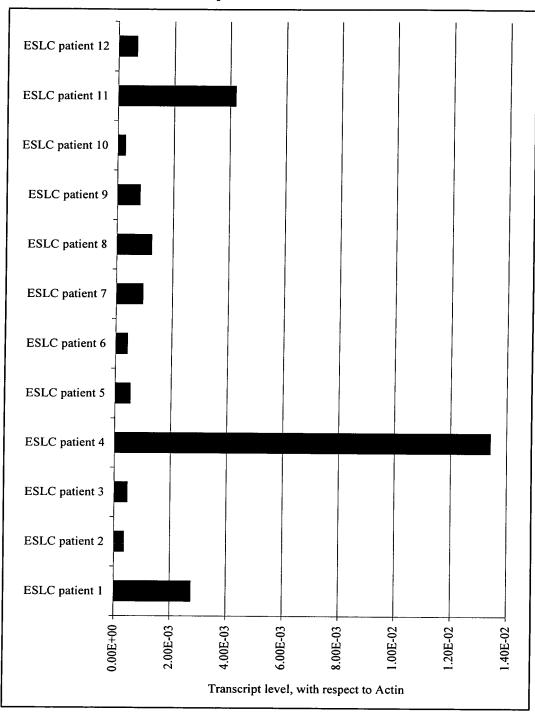


Figure 26: Real-time RT-PCR quantification of CASB7439 transcript on a population of late stage lung cancer (LSLC) patients.

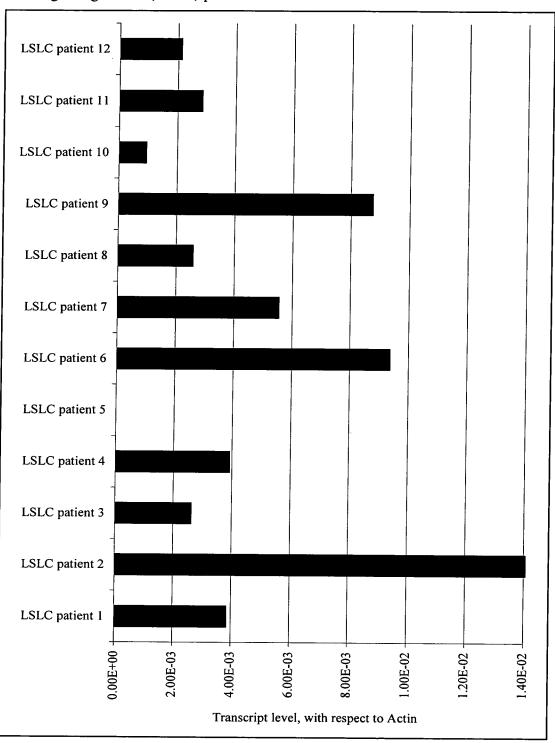


Figure 27: Real-time RT-PCR quantification of CASB7439 transcript on a panel of normal tissues.

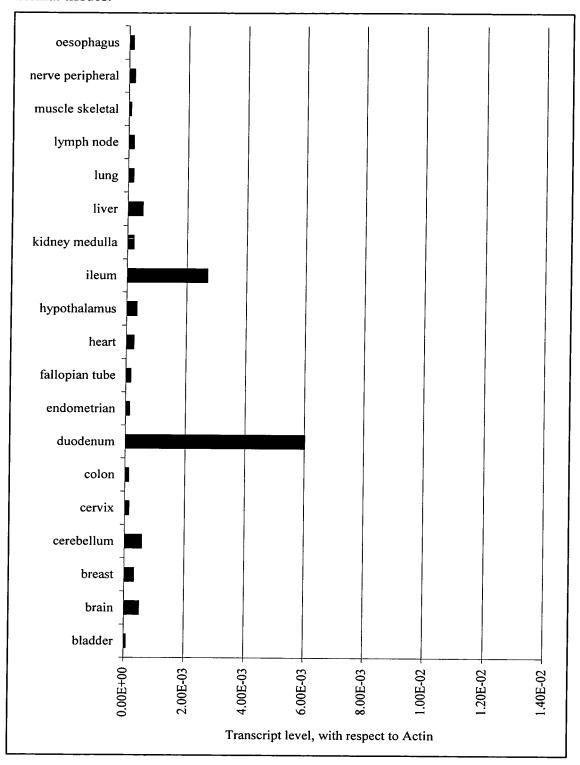


Figure 28: Real-time RT-PCR quantification of CASB7439 transcript on a panel of normal tissues, continued.

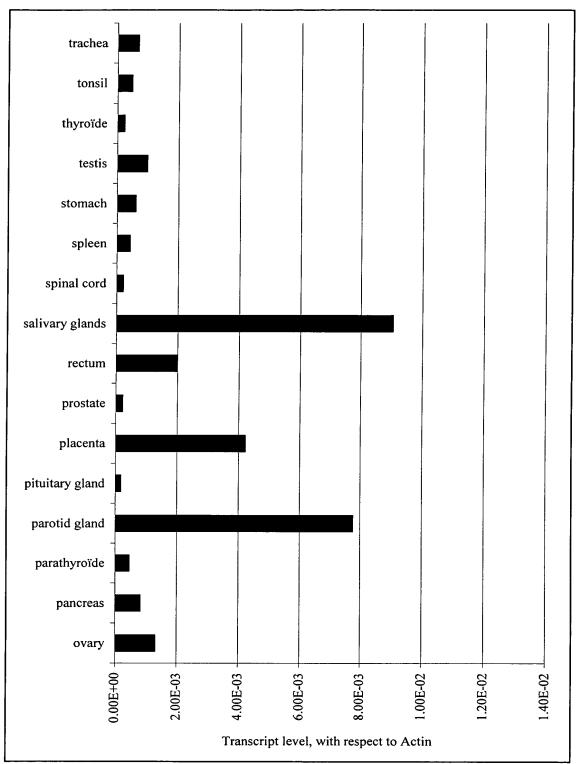


Figure 29: Design of NS1-CASB7439 protein.

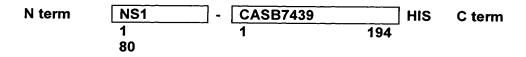


Figure 30: Schematic represention of pMG1 cloning strategy (constructs 1 and 2).

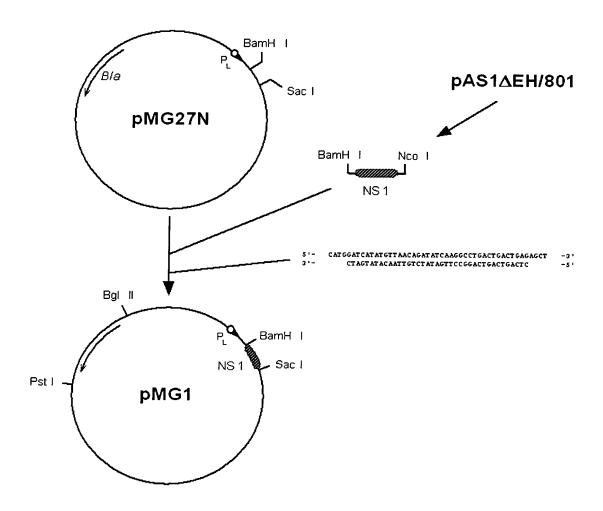


Figure 31: Comparison of Constructs 3 to 5 expression yields. Western blot on CASB7439 constructs 3 to 5 expressed in *E. coli*, revealed with antipeptide SB600 antibody.

	Construct	t 3	Construct 4			Construct 5
1 2 3	4 5	6 }	8 3	lo	AA AZ	A3 A4
### 					•	
_r _r 			\$ ^	. ****	and the second s	

5

Lane 1: MW; Kd

Lane 2: Construct 2, undiluted.

Lane 3: Construct 2, 1/10 dilution.

Lane 4: Construct 2, 1/50 dilution.

Lane 5: Construct 2, 1/100 dilution.

Lane 6: Construct 3, undiluted.

Lane 7: Construct 3, undiluted.

Lane 8: Construct 3, 1/10 dilution.

Lane 9: Construct 3, 1/50 dilution.

Lane 10: Construct 3, 1/100 dilution.

Lane 11: Construct 4, undiluted.

Lane 12: Construct 4, 1/10 dilution.

Lane 13: Construct 4, 1/50 dilution.

Lane 14: Construct 4, 1/100 dilution.

Figure 32: CASB7439 peptide 1 immunization of rabbit SB598: graphical view of ELISA read-outs (Antibody titers relative to CASB7439 peptide 1, straight line; antibody titers relative to KLH carrier, discontinuous line)

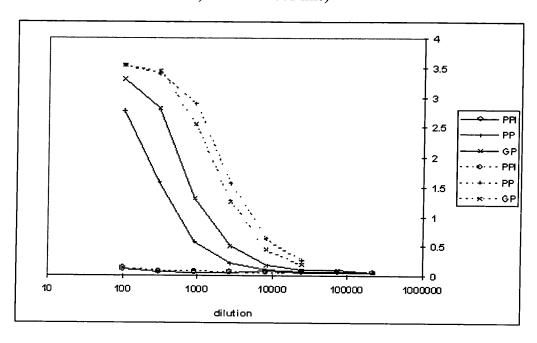


Figure 33: CASB7439 peptide 1 immunization of rabbit SB599: graphical view of ELISA read-outs (Antibody titers relative to CASB7439 peptide 1, straight line; antibody titers relative to KLH carrier, discontinuous line).

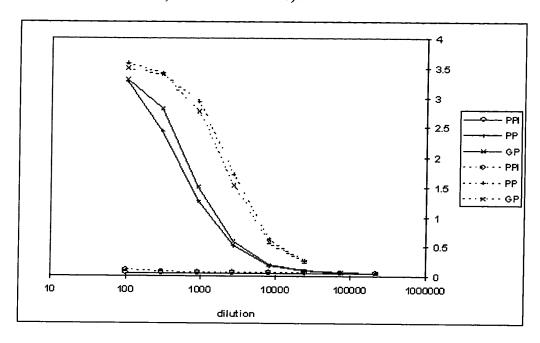


Figure 34: CASB7439 peptide 2 immunization of rabbit SB600: graphical view of ELISA read-outs (Antibody titers relative to CASB7439 peptide 2, straight line; antibody titers relative to KLH carrier, discontinuous line).

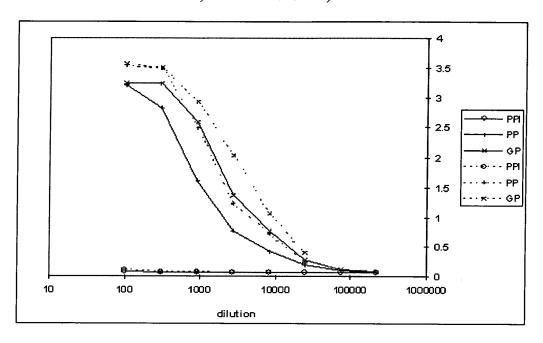


Figure 35: CASB7439 peptide 2 immunization of rabbit SB601: graphical view of ELISA read-outs (Antibody titers relative to CASB7439 peptide 2, straight line; antibody titers relative to KLH carrier, discontinuous line).

